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Genetics, Phenotype, and Natural History of Autosomal Dominant Cyclic Hematopoiesis

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Cyclic hematopoiesis (CH, or cyclic neutropenia) is a rare disease manifested by transient severe neutropenia that recurs approximately every 21 days. The hematologic profile of families with the autosomal dominant form (ADCH) has not been well characterized, and it is unknown if the phenotype is distinct from the more common sporadic congenital or acquired forms of CH. We studied nine ADCH families whose children displayed typical CH blood patterns. Pedigrees confirmed dominant inheritance without evidence of heterogeneity or decreased penetrance; three pedigrees suggested new mutations. Families were Caucasian with exception of one with a Cherokee Native American founder. A wide spectrum of symptom severity, ranging from asymptomatic to life-threatening illness, was observed within families. The phenotype changed with age. Children displayed typical neutrophil cycles with symptoms of mucosal ulceration, lymphadenopathy, and infections. Adults often had fewer and milder symptoms, sometimes accompanied by mild chronic neutropenia without distinct cycles. While CH is commonly described as "benign", four children in three of the nine families died of *Clostridium* or *E. coli* colitis, documenting the need for urgent evaluation of abdominal pain. Misdiagnosis with other neutropenias was common but can be avoided by serial blood counts in index cases. Genetic counseling requires specific histories and complete blood counts in relatives at risk to assess status regardless of symptoms, especially to determine individuals with new mutations. We propose diagnostic criteria for ADCH in affected children and adults. Recombinant human granulocyte colony-stimulating factor treatment resulted in dramatic improvement of neutropenia and morbidity. The differential diagnosis from other forms of familial neutropenia is reviewed.

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KEY WORDS: cyclic hematopoiesis, cyclic neutropenia, familial neutropenia

INTRODUCTION

Cyclic hematopoiesis (CH), also known as cyclic neutropenia, is a rare and distinct disease of neutrophil production. The frequency of this disorder is not well documented, but based upon referral patterns in several large U.S. metropolitan areas, the frequency is estimated to be one per million (Dale, personal communication). Presentations can be sporadic congenital, acquired, or the less common familial form. All forms of CH seem to have in common chronic neutropenia with regular oscillations of neutrophil levels approximately every 21 days [Dale and Hammond, 1988]. The neutrophil nadir lasts 3–7 days with concomitant monocytosis. Neutrophil levels usually oscillate between nadirs of 0–200/mm^3 absolute neutrophil count (ANC), and peaks that rarely exceed 2,000 ANC/mm^3, the lower limit of normal. Marrow examination shows transient "maturation arrest" at the promyelocyte stage preceding each cycle, with subsequent resolution. Because some individuals display cycling of other blood cells with the same periodicity, the name cyclic hematopoiesis more accurately describes this disorder than the common name of cyclic neutropenia. A few adults have been described to stop cycling and develop mild chronic neutropenia [Dale et al., 1993a]. Secondary effects due to severe neutropenia recur during nadirs and include fever, mucosal ulceration, lymphadenopathy, and infections. However, these manifestations may diminish with age [Dale and Hammond, 1988]. Most cases are congenital and two thirds are considered non-familial or sporadic. Acquired cases can present at any age, rarely in association with other blood or im-

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mune disorders, or neoplasms [Chikkappa et al., 1980; Loughran and Hammond, 1986].

The familial form of this disorder appears to be autosomal dominant (ADCH) [(MIM 162800) Online Mendelian Inheritance in Man (OMIM), 1994)] but large pedigree structures and multigenerational blood counts are usually not reported. Although a number of case series described clinical manifestations, most do not distinguish familial and non-familial cases, or pool data from both. Most families of individuals with CH are not evaluated. It has been assumed that individuals with ADCH have a phenotype identical to sporadic CH, but this has never been confirmed. The natural history of the familial disorder also has not been studied.

The actual incidence, gene frequency, and familial blood patterns of ADCH are unclear. Twenty reports describing familial CH from the 1950s to the late 1980s have been reviewed [Dale and Hammond, 1988; Lange, 1983; Morley et al., 1967; Wright et al., 1981]; some mentioned only a family history of neutropenia. Additional CH families have been reported [Bodenstein et al., 1976; Cap and Mikulecky, 1988; Chadwick et al., 1989; Hammond et al., 1989; Inoue et al., 1992; Jonsson and Buchanan, 1991; Tsunogake et al., 1991; see also Methods]. Most reports describe the propositus and sometimes briefly a parent or sib, without characterization of additional relatives. When given, the ethnicity was Caucasian in reports from Europe, the United States, and Australia. Three Japanese families were also included in the above reports. It should be noted that many reports do not list diagnostic criteria, or present insufficient serial counts to confirm the diagnosis of CH; a few present data that do not meet current diagnostic criteria and could be consistent with another disorder. For example, the single African-American family reported [Long et al., 1983] is likely to have a different familial neutropenia, as all ANCs were higher than those seen in CH nadirs, and recurrent periodicity was not documented.

Only one report offers comparisons of several multigenerational families with ADCH. Morley et al. described 19 neutropenic persons in four Australian families diagnosed by the following criteria: neutropenia below 2,000 ANC/mm^3 and oscillating neutropenia in one relative [Morley et al., 1967]. This case series described variable phenotype within families and similar range of manifestations between families, including improved symptoms and chronic neutropenia in a few adults. However, examination of neutrophil patterns suggests, that one, and perhaps two, of the four families did not meet current CH diagnostic criteria. Three other single families have been well described, with cycling demonstrated in childhood, and either cyclic or chronic neutropenia in older generations (families 601, 602, 603 this report, references in Methods).

Owing to the lack of information available on the familial form of CH, we studied a group of families ascertained for our ongoing research to map the ADCH gene by genetic linkage analysis [Palmer et al., 1994]. Our goals were to define the genetics and phenotype of ADCH, including inheritance pattern, penetrance, variable expression, and heterogeneity. In addition, information on detection of cycles, natural history, and morbidity and mortality would be of value in the diagnosis and medical management of this unique form of neutropenia.

METHODS

Fourteen multigenerational families from the United States, Great Britain, and Australia that carried the diagnosis of ADCH were ascertained by referral for evaluation, study participation, or literature review. Some individuals in families CN601-605 were described previously [Andrews et al., 1979; Balazovich et al., 1991; Hammond et al., 1989; Katz et al., 1988; Krance et al., 1982; Lange et al., 1981; Palmer et al., 1994; Parmley et al., 1984; Williams and Jones, 1984; Wright et al., 1994]. If adequate serial ANCs from the propositus were unavailable, serial complete blood counts (CBCs) with manual differential were obtained 2–3 times a week for a minimum of 5 weeks from at least one young neutropenic relative. It was not feasible to obtain serial counts on each affected relative due to reluctance of many subjects and logistics of obtaining numerous blood samples and manual counts. In addition, 18 individuals had subsequently received recombinant human granulocyte colony-stimulating factor, which affects the amplitude and periodicity of cycling [Dale et al., 1993a]. The hematologic diagnostic criteria of CH in each proband consisted of ANC fluctuating between 0 and 2,000 ANC/mm^3 approximately every 21 days, with nadir of 0–200 ANC/mm^3 lasting 3–7 days [Dale and Hammond, 1988]. As peaks often have an irregular shape and amplitude, cycle length was measured from beginning of nadir (first point below 200/mm^3 ANC) of each cycle [Wright et al., 1981]. To detect neutropenia (ANC below 2,000/mm^3) in relatives, at least one neutrophil count was obtained on all available relatives, especially parents of offspring with apparent new mutation; if history or ANC was equivocal additional counts were obtained. All data were obtained prior to recombinant human granulocyte colony-stimulating factor (rhG-CSF) treatment. All available relatives were studied by physical examination (or if unavailable by exam records or physician verification), symptom and medical histories, and family history. Those with neutropenia and/or histories positive for symptoms of CH including fever, lymphadenopathy, mucosal ulcers, early tooth loss, infections, or other manifestations [Dale and Hammond, 1988; Wright et al., 1981], or histories of unclear chronic or recurrent illness requiring medical care, were further evaluated by review of medical records or summaries. All histories, medical records, and questionnaire information included only those periods prior to rhG-CSF treatment.

Relatives determined to be affected by the above data were asked to complete a questionnaire (parents completed for children) to standardize information on signs, symptoms, complications, medical treatment, hospitalizations, and reproductive or other problems. To assess cyclic signs and symptoms that occurred during the nadir, we elected to use patient-reported

frequencies, as this measurement was more objective; subjective severity estimates proved difficult to standardize and record. Symptom frequency was assessed in four categories to simplify patient recording: very frequent (≥9 times a year), frequent (5–8 times a year), sometimes (1–4), and less than once a year.

To assign overall severity designation, objective considerations were made as to number of hospitalizations or procedures, and symptom and complication frequencies reported by questionnaire. Where data were lacking, this was occasionally supplemented by medical records, or clear family history in one individual (family CN614).

RESULTS

Inheritance

Of the original 14 families, 1 had incomplete serial data, and 4 had young propositi with neutrophil oscillations that were irregular or deviated significantly from 21 days; these families were not included in this study. The 9 remaining families had propositi with typical CH blood patterns and additional neutropenic relatives (Fig. 1). Neutropenia was documented in all individuals designated as affected except one, the symptomatic parent of two affected children (presumed obligate carrier CN601 matriarch, deceased without records). Only one person with neutropenia was asymptomatic and lacked a suspicious history (affected CN602 patriarch). A number of individuals in several families were believed by themselves, relatives, and/or physicians to be affected based upon occasional mouth ulcers and infections, tooth loss, and fatigue; these individuals were found to have normal ANCs and lacked the typical periodicity, frequency, and spectrum of symptoms (one had unrelated chronic illness) and were designated unaffected. Available unaffected relatives with normal ANCs and physical findings are indicated in the pedigrees. Of the affected individuals, examination information was available on three-fourths, returned questionnaires on three-fourths, and medical records and serial CBC information on one-half.

All families were of Caucasian origin with the exception of CN601, where the first-generation obligate carrier and spouse were Cherokee Native American. Out of 261 relatives, 67 were determined to be at risk for disease based on dominant inheritance and relationship to known affected propositi and consultants; 43 individuals (64%, vs. expected 50%) were found to

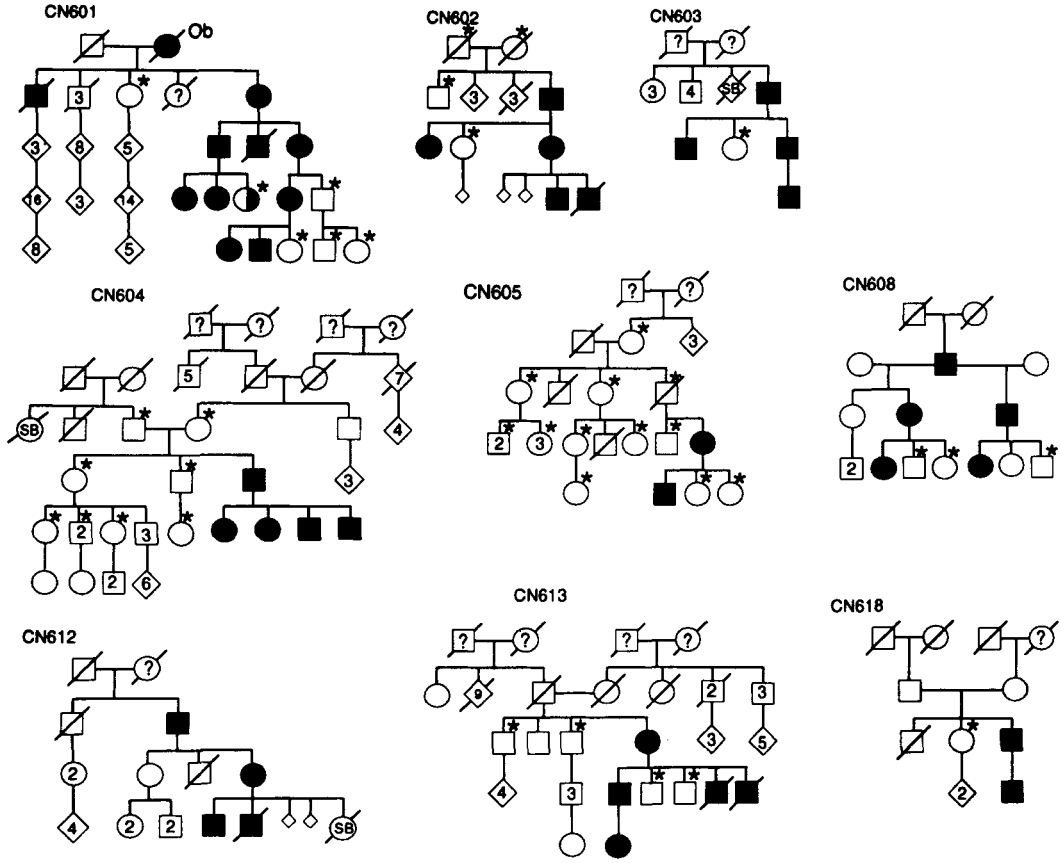

Fig. 1. Pedigrees of ADCH families. Solid symbol, affected with ADCH (confirmed by ANC). Solid symbol with Ob, affected obligate carrier (not confirmed by ANC). Open symbol with *, confirmed unaffected by ANC. Open symbol, unaffected by history (not confirmed by ANC). Half solid symbol, unaffected but acquired ADCH by bone marrow transplant (BMT) [Krance et al. 1982]. SB, stillbirth. ?, status unknown due to inadequate information.

be affected based on neutropenia and symptoms (Fig. 1). Male-to-male transmission occurred in four families, consistent with autosomal dominant inheritance. The sex ratio was 25 males:18 females (1:0.72). There was no reported or observed difference in severity of neutropenia or symptoms between affected males and females. Decreased penetrance was not evident, as all affected sibs had an affected parent, and no unaffected persons had both an affected parent and affected child. Three families (CN602, 603, and 605) presented evidence for new mutations, with single affected child, unaffected parents and sibs, and negative family history. Affected individuals in three families had normal karyotypes (CN601, CN604, and CN605). Normal karyotypes were reported previously in a fourth family [CN602, Lange et al., 1981]. Negative histories of some deceased individuals in older generations could not be confirmed due to unavailable records or absence of blood testing.

Hematologic Patterns of ADCH

The spectrum of ANC cycle variations and patterns can be observed in repeat cycles in individuals, and by intra- and inter-familial comparisons (Figs. 2, 3). The classic CH blood profile is demonstrated by a 26-year-old man with ADCH, with precise neutrophil oscillations every 21 days, and opposite cycling of monocyte levels (Fig. 2A, family CN613). As depicted by the following families, most children had definite periodicity in cycle length but may show more amplitude variability within neutrophil peaks and between cycles. Remarkably similar periodicity of neutrophil nadirs in sibs is displayed in one family (Fig. 2B, ages 5–6, CN604). Another family demonstrated a unique iatrogenic phenomenon (Fig. 3A–D, CN601). Two sibs were diagnosed with ADCH in childhood (Fig. 3B,D, cycles at ages 18–19). Another sib was documented to be unaffected by multiple serial ANCs by age 2, but later acquired acute lymphoblastic leukemia. She was successfully cured but acquired ADCH with a bone marrow transplant from her sib with ADCH [Fig. 3B; Krance et al., 1982]; she continued to show cycling in these serial counts taken 8 years later (Fig. 3C). Their father shows the milder chronic acyclic neutropenia reported in some adults (Fig. 3A, two samplings at age 48). A common cycle length in first cousins was observed (Fig. 3 F–G, ages 7 and 11, CN608), but one child's mother has chronic acyclic neutropenia (Fig. 3E, age 30).

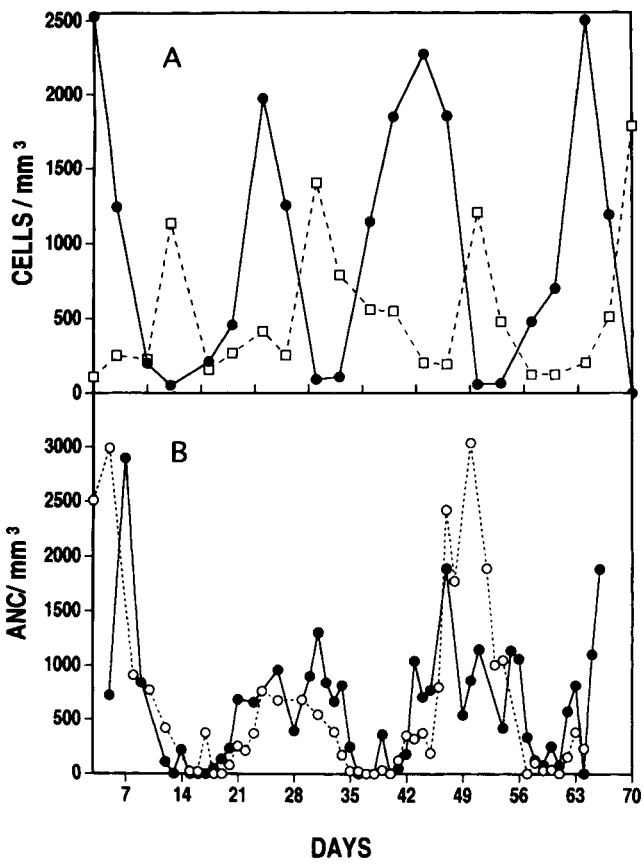

Fig. 2. Patterns of neutrophil cycles illustrating two features of ADCH. **A:** Typical reciprocal fluctuations of neutrophils and monocytes from an individual affected with ADCH (family CN613): Solid line and solid circles, ANC; dashed line and open squares, monocytes. **B:** Similar ANC cycle lengths in two affected siblings (family CN 604), each indicated by separate solid line/solid circle or dotted line/open circle.

Cycles displayed in available serial blood counts are summarized in Table I. Note that of four persons age 30 or older, only one showed cycling. Cycling was displayed by the only person with serial data in the age range of 20–29 (Fig. 2A). All fifteen persons below age 20 displayed cycling, although one individual showed more irregularity when sampled at age 18 (Fig. 3D) than at age 5 [Krance et al., 1982]. Some individuals had occasional peaks that exceeded 2,000, but these rarely lasted longer than 1 day. The observed amplitude variation within cycle peaks in many individuals supported the need for serial neutrophil sampling as frequently as 2–3 times a week, and for 2 cycle lengths, to confirm the cyclic pattern (Figs. 2, 3; data not shown).

Observed cycle lengths ranged from 18 to 24 days, with a typical length of 21 days (Table I). This is in agreement with previously published CH parameters [Dale and Hammond, 1988; Wright et al., 1981]. Nadir (ANC < 200) length was quite variable, ranging between 2.5 days and 9 days. Serial monocyte counts showed opposite cycling in all but one of fifteen individuals (Table I). As some data were previously or locally collected, we were able to trace cycling of other cells in only a few individuals. Of these, suggestions of oscillations of reticulocytes, lymphocytes, or eosinophils were observed in some, but no consistent pattern was detected (Table I). There were insufficient data to determine if distinctive patterns in other cell lines were present in certain families.

Signs, Symptoms, and Complications in Childhood

Characteristic multiple signs and symptoms were displayed in ADCH families (Fig. 4). As many symptoms are shared by unaffected children, we chose to report those frequencies of five or more times a year to minimize overlap with the normal population. Recur-

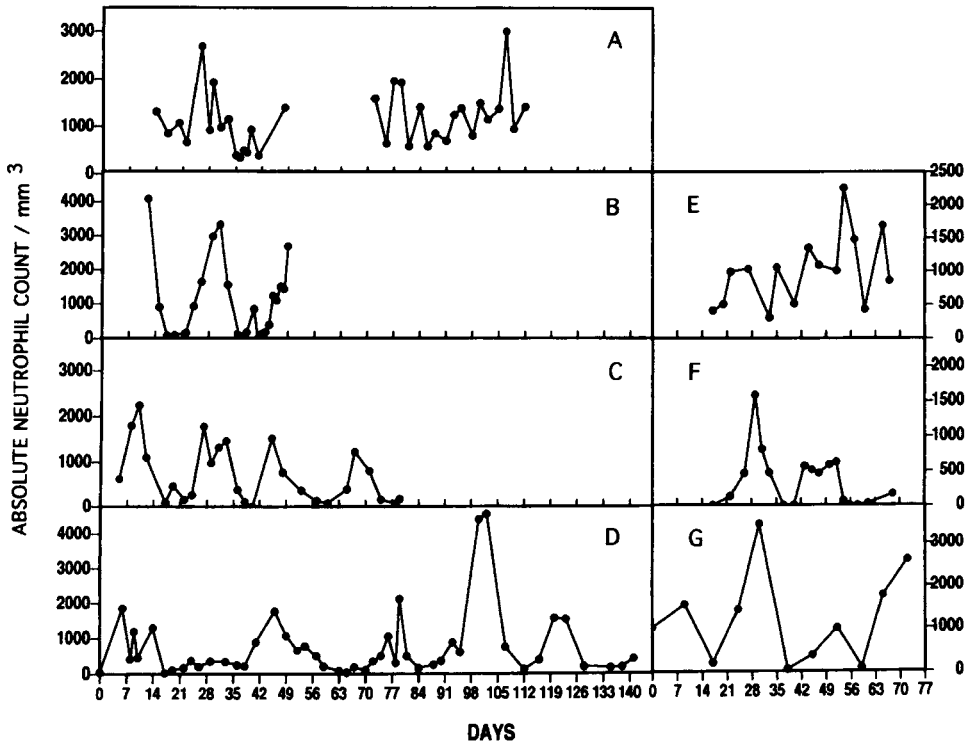

Fig. 3. Intrafamilial and cycle-to-cycle variability of ANC in multiple ADCH family members. Solid symbols, ANC. **A–D:** CN601 family: A, affected father; B, affected daughter, donor of BMT for C; C, unaffected daughter with acquired ADCH by BMT; D, another affected daughter. **E–F:** CN608 family: E, affected mother of F; F, affected daughter of E; **G**, affected niece of E.

ring symptoms were reported by 30 individuals during childhood (below age 18). More than 60% experienced particular symptoms five or more times a year, including oral ulcerations, gingivitis, lymphadenopathy, fever, pharyngitis/tonsillitis, fatigue, or skin infections. More than 30% reported at least five episodes/year of sinusitis, otitis, headache, or depression. Over 20% of children reported the same frequency of bone pain or tooth abscesses; over 10% reported pneumonia, bronchitis, diarrhea, or anal ulcers. Although most symptoms are nonspecific, the frequency of many is undoubtedly increased, and the predictable recurrence of symptom clusters is unique.

Hospitalizations of children were variable and ranged from never to multiple times a year. Owing to incomplete records and nonspecific recall by many frequently hospitalized patients, we were unable to assess precisely the frequency of specific admissions. Common reasons given for admission were rule out sepsis (percent confirmed sepsis unclear); dehydration from fever, emesis, diarrhea, and/or anorexia; cellulitis of limb, face, or perineum; pneumonia; drainage of abscess; necrotizing gingivitis or stomatitis; and rule out acute abdomen (data not shown). Tooth extractions and dental procedures requiring general anesthesia were common. Some urinary tract symptoms were reported, but it is unclear if these were distinguished from urethral and vulvar ulceration. Meningitis, joint infections, and pyelonephritis were rare. No family presented a unique pattern of symptoms and complications. We found it remarkable that we found no instances of serious neonatal infections or sepsis. In fact, many infants were without problems until age 6 to 9 months; this was reported to correspond with weaning from breast milk in only one case. Only a few affected infants were tested at birth, all asymptomatic. However, in one case, cycling clearly began in the first week of life with the first symptoms related to neutropenia (fever and otitis media with poor response to antibiotics) occurring in the second cycle, 3 weeks later. Significant school absence by most children during neutropenic nadirs was reported, due to malaise and fatigue, pain due to ulcerations, and symptoms noted above (data not shown).

Mortality in Childhood due to ADCH

Four deaths occurred during childhood: one 6-year-old boy, one 12-year-old boy, and two brothers, both near age 16. Interestingly, all had affected mothers. All had significant histories of increased symptoms and illnesses. The two unrelated boys had multiple episodes of abdominal pain. All died following an acute illness with abdominal pain and/or emesis rapidly progressing to necrotizing enterocolitis, peritonitis, and *Clostridium* and/or *E. coli* sepsis. One boy had a previous episode with gangrenous terminal ileum [Lange et al., 1981]. Therefore, abdominal pain is the most specific predictor of mortality. However, abdominal pain may be difficult to assess, particularly prior to age 18, as one third of childhood histories demonstrated recurrent abdominal pain, usually with

TABLE I. Variability of Neutrophil Cycles in Individuals With ADCH*

Family	Age	Number of days studied	Cycle range, days	Estim. cycle, days	Typical range ANC	Peak ANC	Other cycles	Nadir, days
601	50	72	No cycles		250–2,000	3,000		
	5/18	200	21–22/17–28, I	22	0–2,200	4,500	m, l	7
	6/19	116	18–24	21	0–2,500	5,000	m, r	6
602	14/16	110	20–21	21	0–2,300	6,000	m, r, u	4
	7	80	20–23	21	0–2,000	3,000	m, u	7.5
	2	80	20,I	20	0–2,000	2,000	m, u	6
603	34/40	124	21–24	23	0–1,800	3,000	m, e	8
	13	160	16–23	20	0–2,500	3,000	u	4
	15	135	18–22	20	0–1,800	2,500	u	7
604	6	60	23–24	24	0–1,900	2,900	m, l	6
	5	60	20–22	21	0–1,900	3,000	m, l	7
605	21	43	21	21	0–2,200	2,500	m, u	5.5
608	30	55	No cycles		300–1,700	2,300		
	11	51	20	20	0–1,700	1,700	None	9
	3/7	113	21–23	22	0–2,000	3,500	m, u	2.5
612	30	50	No cycles		0–1,800	2,400		
	8	50	21	21	0–1,800	1,800	m, u	3.5
613	26	70	20–22	21	0–2,000	2,500	m, u	5
	Newborn	58	18–22	20	0–1,885	3,100	m, u	4, I
618	8	40	21	21	0–1,200	1,200	m, u	9
			Approx. cycles = 21 days					Approx. nadir = 6 days

* n1/n2, sampled at 2 ages; I, indeterminate period; Other cycling observed: m, monocyte; e, eosinophil; l, lymphocyte; r, reticulocyte; u, other cell counts unavailable.

nausea and vomiting. Five children reported 9 or more episodes/year, one reported 5–8 episodes/year, and four children reported 1–4 episodes/year.

The records and histories of the deceased boys were examined for any other possible predictor of mortality. The brothers were below the 5th centile in height by parents' report. Out of self-reported heights, only 2 additional persons out of 29 had height below the 5th centile for age; these were severely symptomatic brothers of the two unrelated deceased boys. One had successful resection of intestine due to infection at age 20; the other is doing well on rhG-CSF treatment. No case of short stature was predicted based on parental heights. Although the clustering of these six cases in three families might suggest genetic heterogeneity, individuals in other families showed similar abdominal symptoms (without sequelae) or other severe complications. All nine families displayed a range of severity, and one family in which a death occurred included the asymptomatic individual (data not shown). Based on clinical signs there is no evidence for locus heterogeneity.

Signs, Symptoms, and Complications in Adults

Symptom frequencies of adults suggest improvement with age when compared to that of children, supporting previous anecdotal reports for both sporadic and familial CH (Fig. 4). Sinusitis, headache, and bone pain are more common, although oral ulcers, fatigue, and gingivitis are still prominent. Less common are skin infections, fever, lymphadenopathy, and pharyngitis. Each adult was asked to report symptom frequencies of both childhood and adulthood. Comparing those for each individual, and combined with additional records, 13 of 19 experienced marked improvement of symptoms and infections, 5 reported little or no change (including the only asymptomatic person), and 1 person had significant worsening with more frequent hospitalizations for infections and dehydration, and prolonged work absence. However, for most adults hospitalizations were far less frequent, and work absence was uncommon (data not shown).

A prevalent phenomenon in all disorders of chronic neutropenia is early permanent tooth loss resulting from chronic gingivitis, tooth abscesses, and alveolar bone loss. Of 27 adults with ADCH (most younger than age 50, many decades younger), 13 reported tooth loss prior to age 30, with 7 of these edentulous, including 2 in their teens. Four others lost teeth in their early 30s or at a "young" age. As some adults are not yet age 30, these results may underestimate the frequency of early permanent tooth loss.

Reproductive Issues and Assessment of Associated Anomalies

Eight affected women reported 25 pregnancies, with complications in 9. One woman had three premature deliveries before 32 weeks of gestation; one child has cerebral palsy (unaffected with ADCH). Another woman had a severe episiotomy infection with recurrent rectovaginal fistulas, Cesarean section wound infection, a stillbirth at 20–30 weeks, a ruptured ectopic pregnancy, and possibly another early loss. One woman reported two pregnancy losses, and another reported one loss (details unknown). Another woman had "difficulty recovering" following childbirth (details not given). Both worsening and improvement of symptoms during pregnancy were reported. Correlation of complications to neutrophil nadirs is unknown. In comparison, no problems were reported by mothers when 10 affected males fathered 23 pregnancies.

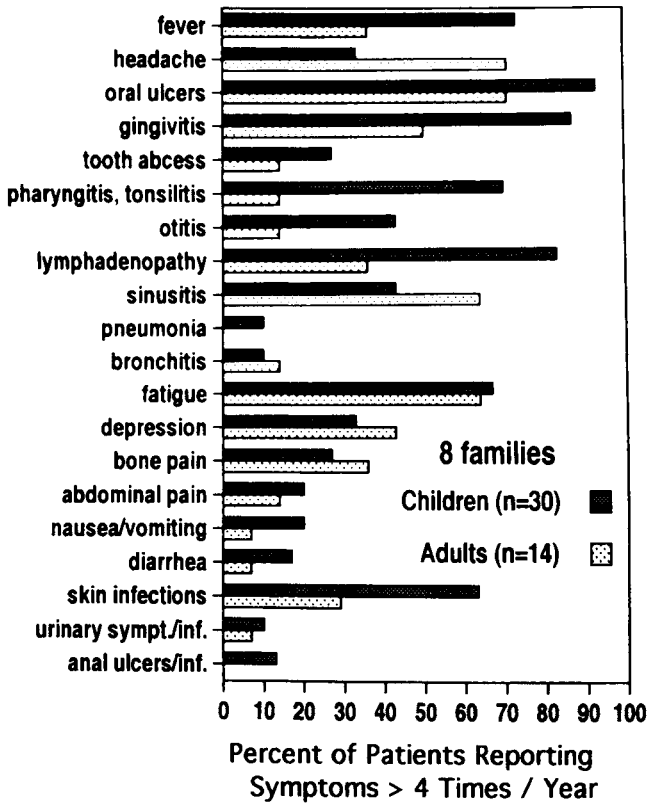

Fig. 4. Frequent signs and symptoms of ADCH. Bars represent percent of patients reporting each category five or more times a year. Dark bars, children under age 18; Light bars, adults age 18 and older.

We investigated the possibility of any anomalies associated with ADCH. One case of cleft palate was found. No other anomalies were reported or noted on exam or in records. Owing to the association of pigmentation or ray defects seen in other congenital blood disorders, these questions were specifically asked in interviews and on questionnaires concerning unavailable relatives. Three individuals with a "white spot" were reported by two families without further details. Based on these findings, there is no information to suggest an association between ADCH and any anomaly.

Natural History of ADCH

Of 26 affected individuals from nine families who survived into adulthood, 4 adults are deceased. One woman, age 53, died of unknown cause over 50 years ago. One man, age 80, died of a stroke. Another man, age 64, had pneumonia at the time of death following complications of heart surgery, but suffered from previous stroke and pneumonia seemed to be a complicating rather than precipitating factor. Only one person died of malignancy, a man deceased at age 48 from renal cancer. Of 22 living adults at time of data collection, one = age 80, two = ages 70–79, one = age 60–69, four = ages 50–59, and the remainder were under age 50. Additional illness reported were one case each of hypertension, colon cancer, arthritis, and thymoma. Four cases of transient anemia in women were reported. There were no cases of other blood disorders or leukemias, and this small sample size does not suggest a specific association with malignancy.

DISCUSSION

Autosomal Dominant Cyclic Hematopoiesis

We present research on nine families that supports the presence of an autosomal dominant gene, ADCH, responsible for their familial CH. The distinct periodic phenotype may later become chronic, as demonstrated by the presence of intrafamilial cases of periodic neutropenia and symptoms in all affected children, and chronic neutropenia and symptom amelioration in many affected adults. Both phenotypes occurred in the same individual (family CN602) [Lange et al., 1981]. No evidence was found to suggest decreased penetrance in any family.

The nine ADCH families displayed no evidence for locus or allelic heterogeneity in blood or symptom phenotypes. However, three additional families had irregular or shorter cycles, with one woman suffering from unusually severe pneumonias and pregnancy complications (data not shown). It is unclear whether these "atypical" families are affected with ADCH or a similar disease. Acquired CH, with its atypical lymphocytes or other white cell abnormalities, is not familial [Ferrero et al., 1993]. Therefore, the various acquired presentations may not be caused by a primary defect in the germline. A clonal mutation or chromosomal rearrangement arising in marrow cannot be ruled out. In one interesting family that defies classification, CH was acquired by teenaged twins several years apart [Chusid et al., 1986].

Each ADCH propositus presented with at least moderate symptoms and received medical care on multiple occasions. Ascertainment bias seemed to play a role in the failure to identify milder ADCH in adult relatives of propositi in six families. This was demonstrated by the asymptomatic neutropenic father of affected children in one family, and several other families with delayed recognition of familial disease (data not shown).

The phenotype of ADCH seems indistinguishable from that of sporadic congenital CH. This suggests the possibility that some sporadic cases may represent ADCH either as new mutations, as suggested by apparent new cases in one third of the ADCH families, or as unrecognized familial cases. The second possibility was demonstrated by the observed ascertainment bias.

Differential Diagnosis

The differential diagnosis of ADCH includes other familial congenital neutropenias. Although some have distinct pathology in the hematology literature, from a genetic standpoint they seem to represent poorly defined clinical groups. There appears to be phenotypic overlap between groups, and variable expressivity and possibly genetic heterogeneity within each. As with ADCH, few families have been studied, so intra- and inter-familial variation is unclear. Few cases are reported with serial cell counts, so the possibility exists that some might represent ADCH. Each diagnosis was previously held by one or more families in our study,

and many ADCH individuals, if considered without family or cycle data, might appear to fit into one of these categories:

1) Kostmann syndrome [(MIM 202700) OMIM, 1996]: This autosomal recessive severe congenital neutropenia (SCN) was originally described in inbred Swedish kindreds [Iselius et al., 1984; Kostmann, 1975]. Children have profound neutropenia ($<500/mm^3$ ANC) with persistent marrow arrest, and rarely survived prior to modern treatment. This diagnosis is commonly misused without regard to inheritance or ANC pattern.

2) Benign familial neutropenia [(MIM 162700) OMIN, 1994]: This mild autosomal dominant congenital neutropenia has fewer symptoms (mostly gingivitis). ANCs range between 1,000 and $2,000/mm^3$. It is more common in Middle Eastern, Yemenite Jewish, and African ethnic groups [Ash et al., 1986; Shoenfeld et al., 1988].

3) Congenital neutropenias: Other families with neutropenia clearly do not fit the above two groups. In addition to ADCH, we propose the following categories, which may be genetically distinct groups:

a) Autosomal dominant severe congenital neutropenia: We have observed one father-child pair with a non-cyclic, relatively severe neutropenia.
b) Autosomal dominant congenital neutropenia (intermediate severity): In addition to the atypical ADCH families, we have observed non-cyclic families with ANC and symptoms intermediate to that of SCN and benign neutropenia (data not shown). Some report improvement with age. Other families are found in the literature [Falk et al., 1976; Joyce et al., 1977; Kirstila et al., 1993].
c) Autosomal recessive congenital neutropenia (intermediate severity): Sibs have a less severe course than Kostmann syndrome, although the lack of parental data does not rule out b) above [Koren et al., 1989; Jonsson and Buchanan, 1991].

In any case, relatives must be evaluated, so that patterns of inheritance may indicate recurrence risk even if the type of neutropenia cannot be differentiated.

Other genetic diseases with neutropenia can be ruled out by distinctive findings, such as Barth syndrome, glycogen storage disease type Ib, Shwachman syndrome, reticular dysgenesis, cartilage- hair hypoplasia, Chediak-Higashi albinism, Fanconi anemia, and others [OMIM, 1994; OMIM, 1995; OMIM, 1996]. Other disorders that share manifestations with ADCH but lack neutropenia include autosomal dominant cyclic thrombocytopenia, familial Mediterranean fever, PFAPA syndrome (periodic fever, adenopathy, pharyngitis, and aphthous ulcers), and Behcet disease [Marshall et al., 1987; OMIM, 1994; OMIM, 1995].

Molecular Basis of ADCH

Despite extensive studies of marrow precursors, the cause of the maturation disturbance in CH/ADCH is unknown. Using a genetic linkage mapping approach, we have significant data to exclude the ADCH gene from a location within the major hematopoietic regulatory gene cluster on chromosome 5q [Palmer et al., 1994]. This cluster is comprised of numerous genes encoding cytokines and their receptors, several of which play a role in granulopoiesis such as GM-CSF, M-CSF receptor, IL3, IL4, and IL5. Although a mutation in the G-CSF receptor has been reported in one case of SCN [Dong et al., 1994], our recent genetic studies indicate that neither this receptor nor its ligand, G-CSF, are linked to the ADCH locus (Palmer et al., unpublished). Therefore, at this time, molecular diagnosis of this disorder is not possible.

Medical Aspects of ADCH

CH has long been considered to be benign, and "benign cyclic neutropenia" or "benign neutropenia" was found on medical and autopsy records of ADCH families. Although most infections are not life-threatening, even transient neutropenia below $200/mm^3$ can predispose individuals to serious infection. Most children and some adults had the lifestyle of those with significant chronic disease. With known mortality, the term "benign" is not appropriate for either familial or sporadic forms of the disorder.

The results of this study serve to alert physicians and families to the pattern of mortality and serious morbidity in children and young adults. As stated previously, the most specific predictor of mortality is acute abdominal pain or emesis, particularly with a prodrome suggestive of gastroenteritis. In addition to the four deaths reported here, one other has been reported in ADCH [Jonsson and Buchanan, 1991], as well as a number of sporadic cases [Geelhoed et al., 1973; reviewed by Hopkins and Kushner, 1983]. All episodes of abdominal pain should be clinically followed to rule out acute abdomen, keeping in mind that the ANC is not a useful indicator of infection. Clinicians and families should be alert to toxic signs, and the possibility of rapid progression to septic shock or bowel necrosis. If antibiotics are indicated, coverage for *Clostridium* and other coliforms should be included [Hopkins and Kushner, 1983]. Continuity of care is essential for heightened awareness of intestinal and pregnancy complications. Although the observed lack of neonatal infection is intriguing, the increased susceptibility of even normal infants to sepsis dictates careful postnatal monitoring of infants at risk for ADCH.

RhG-CSF has become the indicated treatment for CH and ADCH with moderate to severe symptoms [Bonilla et al., 1994; Dale et al., 1993b; Hammond et al., 1989]. Familial cases respond in a manner similar to sporadic CH, with increase in ANC, shortened cycles, and amelioration of symptoms [Hammond et al., 1989; Dale, unpublished]. Dramatic increases in feeling of well-being and school attendance were reported by most families (data not shown), as with sporadic cases [Jones et al., 1993]. Although clinical trials to date have indicated few adverse affects in rhG-CSF treatment of ADCH or CH, the outcome of decades of treatment is not known. At this time beneficial effects seem to outweigh any unknown risks of treatment. However, continued monitoring is warranted, as significant findings of apparent

development of monosomy 7, myelodysplasia, or leukemia have been reported in some individuals undergoing rhG-CSF treatment for other congenital neutropenias [Bonilla et al., 1994; Dong et al., 1994; Imashuku et al., 1994]. Whether this association is related to an underlying predisposition specific to those disorders, or perhaps a clonal selection of an undetected pre-existing mosaicism, versus coincidence unrelated to treatment, is unknown.

Diagnostic Criteria

Diagnosis of ADCH can be achieved by individual and family histories along with confirming manual ANCs. We warn against relying on history alone, as the asymptomatic affected and "symptomatic" unaffected individuals illustrated. Although ADCH can be diagnosed by ANCs alone, histories can identify relatives at risk, and those who need additional ANCs, experience abdominal pain, or may benefit from rhG-CSF treatment. We propose the following Evaluation Protocol for ADCH:

1) Construct a pedigree, and ascertain on each individual:
   a) history of neutropenia or blood disorder;
   b) records for previous CBCs, suggestive hospitalizations or procedures;
   c) frequency of signs and symptoms as in Figure 4; dental and pregnancy histories.

2) For the propositus (preferably a child), obtain manual CBCs 2–3 times/week for >6 weeks.

3) After diagnosis of propositus, all first degree relatives (and others with suggestive histories) require up to four weekly CBCs until ANC <2,000/mm^3 is documented in more than one sample. (Although it was not feasible to sample this frequently on all relatives in our study, the presence of equivocal counts dictates more rigorous evaluations for counseling.)

4) Diagnosis of ADCH should meet the following Minimal Diagnostic Criteria:
   a) The propositus should display typical cycles with nadirs of 0–200/mm^3 ANC recurring approximately every 21 days, other ANC usually <2,000/mm^3. Opposite monocyte cycling is usually observed; other cells may cycle. (Adults with acyclic neutropenia require confirmation of typical cycles in a relative.)
   b) At least one first degree relative (if unavailable, another relative) must display ANC below 2,000/mm^3 in > two samples. If serial counts are available, all affected children should display cycling.

Obligate non-penetrant individuals or neutropenic children who do not display typical ANC patterns should be investigated further. If the above criteria are met, neutropenia symptoms are longstanding, and no other manifestations suggesting another blood disease are present (such as blasts, anemia, or indications of leukemia), then bone marrow examination is not indicated for diagnosis. A hematologist should be consulted as soon as possible in any event if any blood abnormality is detected. Serial counts in other relatives are not necessary unless severe infections occur. Serial counts, as well as bone marrow with cytogenetic evaluation, are recommended before initiating rhG-CSF treatment.

We recommend the following Evaluation and Counseling of Sporadic Cases:

1) Evaluate propositus and relatives as outlined in the "Evaluation Protocol for ADCH" above.

2) Affected individuals with negative family evaluation should be counseled that they may carry a new mutation, and each future child may have up to a 50% risk to inherit the ADCH gene. However, the actual a priori risk to offspring is unclear, since reproductive histories of individuals with sporadic CH have not been studied. It is unknown what proportion might carry germline versus somatic marrow mutations, or CH due to another cause. The recent establishment of an international registry for all forms of congenital neutropenia by one of the authors (D.C.D.) may facilitate future study of this and other aspects of CH.

This clinical study of ADCH provides diagnostic, management, and counseling information for geneticists, other clinicians, and families. Identification of the gene responsible for ADCH will hopefully facilitate diagnosis, treatment, and ultimately gene therapy, of this and other marrow disorders.

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